

## Adulteration of butterfat: validity of Reichert-Meissl, Polenske and iodine values

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### RESUMEN

#### Adulteración de grasa de manteca: validez de los índices de Reichert-Meissl, Polenske e iodo.

Durante las estaciones de invierno y verano se obtuvo grasa de leche de búfalo. Sebo de vaca (grasa del corazón) se adquirió del mercado local. El sebo de vaca se añadió a la grasa de leche de verano en las relaciones 5%, 10%, 15% y 50% w/w. Se aplicó la cromatografía en capa fina para obtener la fracción de triglicéridos de cada muestra obtenida. La cromatografía gaseosa se usó para identificar el contenido en ácidos grasos de la grasa de leche de invierno, verano y las mezclas. También se determinaron para cada muestra los índices de Reichert-Meissl (RM), Polenske (P) e iodo. Se concluyó que los (RM) de la grasa de leche de invierno y de verano fueron  $25.52 \pm 0.511$  y  $28.74 \pm 0.568$  respectivamente. Los índices (P) y de iodo fueron  $1.94 \pm 0.162$ ,  $2.53 \pm 0.146$  y  $42.1 \pm 0.85$ ,  $30.33 \pm 0.839$  en los mismos aspectos. La adición de sebo de vaca a la grasa de leche en las relaciones 5% y 10% no afectó los índices (RM) y (P) de las mezclas resultantes. La presencia del 15% de sebo de vaca en la grasa de leche afectó ligeramente los índices (RM), (P) e iodo. La adición del 50% de sebo de vaca a la grasa de leche sacó los valores de (RM) y (P) del rango normal, permaneciendo dentro del mismo el índice de iodo. Se observó también que la adición de sebo de vaca en las relaciones 5%, 10% y 15% a la grasa de leche de verano, mejoró el comportamiento de los ácidos grasos de la grasa de leche, mientras que el 50% de sebo de vaca en la grasa de leche, disminuyó todos los ácidos grasos de cadena corta. La determinación de los índices (RM), (P) e iodo para detectar sebo de vaca como adulterante en grasa de leche pura no es suficiente al 5% y 10% de sebo de vaca en la grasa de leche.

**PALABRAS-CLAVE:** Adulteración (detección) – Grasa de leche – Sebo de vaca.

### SUMMARY

#### Adulteration of butterfat: validity of Reichert-Meissl, Polenske and iodine values.

Buffaloe's milkfat was obtained during the winter and the summer seasons. The beef tallow (Heart fat) was purchased from the local market. Beef tallow was added to the summer milkfat in the ratios 5%, 10%, 15% and 50% w/w. Thin layer chromatography was applied to obtain the triglycerides fraction of each sample obtained. Gas chromatography was used to identify the fatty acid content of the winter, summer and the admixtures. Reichert-Meissl (RM), Polenske (P) and iodine values were also determined for each sample. It was concluded that the (RM) of the winter and the summer milkfat were  $25.52 \pm 0.511$  and  $28.74 \pm 0.568$ , respectively. The (P) and iodine values were

$1.94 \pm 0.162$ ,  $2.53 \pm 0.146$  and  $42.1 \pm 0.85$ ,  $30.33 \pm 0.839$  in the same respect. The addition of beef tallow to the milkfat in the ratios 5% and 10% did not affect the (RM) and (P) values of the resultant admixtures. The presence of 15% beef tallow in milkfat slightly affected the (RM), (P) and iodine values. The addition of 50% beef tallow to the milkfat got the values of (RM) and (P) out of the normal range. The iodine values was still within the normal range of milkfat. It was also observed that the addition of beef tallow in the ratios 5%, 10% and 15% to summer milkfat, improved the fatty acid pattern of the milk fat, while the 50% beef tallow in milkfat, decreased all the short chain fatty acids. The determination of the (RM), (P) and iodine values to detect the beef tallow as an adulterant in pure milkfat is not sufficient at 5% and 10% beef tallow in milkfat.

**KEY-WORDS:** Adulteration (detection) – Beef tallow – Milkfat.

### 1. INTRODUCTION

Adulteration of butterfat with competitive fats had been studied by many investigators. The common methods of detecting such adulteration has consisted in determining the fatty acid composition of the fat obtained from the suspected product (1) (2). However, the microscopic appearance and the melting point of a fat sterol acetate could greatly assist in detecting the vegetable fat in butter fat (3). Concerning the trans isomer content, the genuiness of a suspected sample could be proved by measuring the trans isomer content of such sample. The presence of the trans isomer in a milkfat sample is a sufficient evidence to the presence of hydrogenated oil (4). In other study to detect the adulteration in butterfat, it was found that the  $\beta$ -sitosterol is the predominant sterol of the cottonseed oil, while the cholesterol was the principal sterol of the beef tallow. The beef tallow could not be detected in butterfat up to 6% beef tallow in butterfat, while cottonseed oil was easily detected even at 2% cottonseed oil in butterfat (5). Fatty acid percentage esterified in the 2-position of the triglycerides of milkfat and beef tallow was calculated, using gas chromatography. The presence of 2%, 4% and 6% beef tallow in the milkfat was detected (6). However, many investigators determined certain fat constant values to detect the adulteration in a suspected fat, which were, the butyric

acid content (7). Reichart-Meissl and Polenske values (8), gas chromatographic analysis of two fractions obtained by fractional crystallization (9) and fatty acids ratio of authentic and adulterated milkfat (10).

The author attempts in this work, to evaluate the validity of some common determinations to detect the adulteration of butterfat with other animal fat.

## 2. MATERIALS AND METHODS

### - Source of samples and sampling

Buffaloe's milk samples were collected during the winter as well as the summer seasons, from the herd of Sakha Experimental Station, Animal Production Research Institute. The animal were mainly fed Egyptian clover and concentrate mixture during the winter season, while it fed rice hay and concentrate mixture during the summer season. The sampling was repeated 5 times in both seasons. Milk fat was extracted from each sample after centrifuging, by melting and the fat was filtered. Beef tallow (Heart fat) was purchased from the local (slaughter house). The (chloroform methanol 2-1 v/v) system was applied to extract the fat from tissue. Solvents were evaporated using a rotary evaporator.

The milkfat of winter season was taken as a control due to the balanced plan of nutrition during the winter season. The milkfat of the summer season was mixed with the beef tallow in the ratios 5%, 10%, 15% and 50% w/w beef tallow in milkfat. The obtained admixtures were melted separately to obtain good homogeneity, and then, a proper amount was taken from each sample for analysis.

### - Separation of the samples triglycerides

A preparative thin layer chromatography (TLC) was applied to separate the triglyceride fraction of each sample (11).

### - Methylation

Triglyceride fatty acids were converted to the corresponding methyl esters using methanol, zinc chloride and zinc dust as a catalyst (12).

### - Chromatographic analysis

The determination of the resulted fatty acids methyl esters was carried out using a gas chromatograph, type Hewlett-Packard 5840A, with double flame ionization detector and with multilevel temperature programmer provided with a HP-5840A terminal. Column used was a 6 feet stainless steel packed with 10% UCW 928 on chromosorb W. AM, DMCS treated, 80-100 mesh (Hewlett Packard). Carrier gas used was nitrogen. The conditions were as follows: length of

column, 6 feet, with internal diameter 1/8 inc. Programming temperature 140-300°C, 8°C/min. Detector temperature 300°C, injection port temperature 230°C. Carrier gas flow rate ( $N_2$ ) 40 ml/min. Hydrogen flow rate 40 ml/min. Air flow rate 300 ml/min.

### - Reichert, Polenske and Iodine determinations

The Reichert, Polenske and Iodine values were determined for each sample as described by Bolton and Rvis (13).

## 3. RESULTS AND DISCUSSION

Present data shows the fatty acid composition as well as some constants of winter and summer buffaloe's milkfat. It obvious that both of fats had the same fatty acid pattern. On one hand, the winter milkfat characterized with a high percentage of  $C_{18}$  unsaturated and low levels of short chain fatty acids. On the other hand, the summer milkfat had higher levels of saturated fatty acids, and short chain fatty acids, than the one found in the pattern of winter milkfat. However, this discrepancy between both fats in their fatty acid levels was due to the effect of the season of the year and consequently the plan of nutrition (14) (15) (16). From the same table, the iodine value was found to be higher in winter milkfat than that found in the summer milkfat. The iodine value ranged from  $30.33 \pm 0.839$  in summer milkfat to  $42.10 \pm 0.850$  in winter milkfat (17). The same data contained the Reichert-Meissl (RM) and Polenske value (P). It is clear that the (RM) ranged from  $25.52 \pm 0.511$  in the winter to  $28.74 \pm 0.568$  in the summer, while the (P) ranged from  $1.94 \pm 0.162$  to  $2.53 \pm 0.146$ , respectively. The previous findings were expected because the summer milkfat contained higher levels of  $C_4$  and  $C_6$  acids than the one found in winter milkfat (18) (19). The fatty acid pattern of beef tallow, also in the table, revealed high contents of  $C_{18:0}$  and  $C_{18:1}$  acids. However, these acids besides  $C_{16}$  acid are found to be characteristic for the beef tallow. The iodine values was 54.55.

The admixtures of 5%, 10%, 15% and 50% beef tallow in the summer butterfat are also reported in the Table. The fatty acid content of the admixtures revealed that the fatty acids  $C_4$ - $C_{14}$  were decreased as the added beef tallow increased. On the other hand,  $C_{18}$  acids were increased by the addition of beef tallow to the summer milkfat. These observations are due to the absence of the short chain fatty acids, and the higher content of the  $C_{18}$  acids of the beef tallow (19). The effect of the addition of beef tallow to the milkfat on the iodine, (RM) and (P) values are shown in the table. Several investigators reported that the iodine, (RM) and (P) values ranged from 29.1-41.1, 24.24-33.55 and 1.92-2.95, respectively (20) (21). From the previous results it can be concluded that the addition

Table

Fatty acid composition of buffaloes' milkfat during winter and summer seasons, and of beef tallow and admixtures with beef tallow including the IV, RM and P values.

	Winter milkfat (Dec. - Apr.*) I					Summer milkfat (June - Oct.*) II					Beer tallow III	III+I	III+I	III+I	III+I
	Max.	Min.	Mean	S.E.	S.D.	Max.	Min.	Mean	S.E.	S.D.		5%	10%	15%	50%
C <sub>4</sub>	3.58	2.11	3.00	0.247	0.552	3.71	3.14	3.50	0.105	0.234	-	3.35	3.18	2.59	1.66
C <sub>6</sub>	1.30	0.99	1.40	0.165	0.370	2.17	1.39	1.90	0.134	0.300	-	1.85	1.68	1.31	0.85
C <sub>8</sub>	1.30	0.80	1.00	0.085	0.190	1.83	0.82	1.31	0.211	0.472	-	1.26	1.19	1.10	0.65
C <sub>10</sub>	2.07	1.60	1.90	0.087	0.194	2.93	2.17	2.50	0.151	0.337	0.15	2.40	2.27	2.10	1.10
C <sub>12</sub>	3.80	2.00	2.50	0.172	0.385	4.16	2.56	3.22	0.347	0.775	0.18	3.15	2.88	2.61	1.52
C <sub>14</sub>	11.00	9.69	10.10	0.239	0.535	14.92	13.11	14.35	0.339	0.759	4.83	14.98	13.35	12.73	11.10
C <sub>14:1</sub>	3.11	1.13	2.00	0.342	0.764	1.76	1.33	1.45	0.079	0.178	0.98	1.27	1.61	1.60	0.80
C <sub>16</sub>	29.61	27.98	28.90	0.307	0.686	36.16	32.77	34.55	0.938	2.097	26.15	33.16	31.90	30.96	30.71
C <sub>16:1</sub>	6.60	2.00	4.30	0.963	0.153	4.41	3.00	3.60	0.247	0.552	2.80	3.41	3.32	3.44	2.82
C <sub>18</sub>	10.98	9.11	10.20	0.345	0.771	9.17	6.63	8.40	0.472	1.056	16.32	8.86	9.46	9.89	12.11
C <sub>18:1</sub>	35.87	30.72	32.80	0.856	1.915	26.74	22.11	24.51	0.921	2.056	45.82	25.60	28.40	30.88	35.51
C <sub>18:2</sub>	3.21	1.00	1.90	0.413	0.923	1.35	Trace	0.71	0.295	0.659	1.04	0.71	0.76	0.79	0.77
C <sub>18:3</sub>	2.01	Trace	0.80	0.491	1.098	Trace	Trace	Trace	-	-	1.73	Trace	Trace	Trace	Trace
I.V.	45.30	40.44	42.10	0.850	1.897	32.58	28.08	30.33	0.839	1.877	54.55	31.12	34.52	36.64	41.46
R.M.	27.03	23.98	25.52	0.511	1.143	30.80	27.57	28.74	0.568	1.271	-	27.57	25.47	22.31	13.57
P	2.12	1.57	1.94	0.162	0.362	2.97	2.05	2.53	0.146	0.327	-	2.33	1.91	1.65	0.99

\* Ration was alfalfa + concentrates

\*\* Ration was concentrates + rice hay

I.V. : Iodine value

R.M. : Riechert-Meissl

P: Polenske value

of beef tallow to the summer milkfat did not affect the iodine value even at 50% beef tallow in milkfat. The ratios 5% and 10% beef tallow in milkfat did not affect the (RM) and (P) values being within the range of the pure milkfat, analysed or reported. The percent of 15% beef tallow in milkfat slightly affected the iodine, (RM)

and (P) values. The addition of 50% beef tallow in milkfat got the values of (RM) and (P) out of normal range (1.57-2.12) of the winter milkfat. On the other hand, the addition of 5%, 10%, 15% beef tallow in milkfat improved the fatty acid pattern of the summer milkfat, while the 50% decreased all the short chain

beef tallow as adulterant in pure milkfat is not sufficient at 5% and 10% beef tallow in milkfat.

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